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(54) Title: **HOMOGENEOUS AND HETEROGENEOUS ASSAY METHODS USING NUCLEATION CENTERS AND NOVEL COVALENT CHEMISTRIES FOR THE RAPID MEASUREMENT OF PHOSPHORYLATION AND DE-PHOSPHORYLATION**

(57) Abstract: Novel chemistries, methods and assays are described for the rapid detection and measurement of phosphorylation or de-phosphorylation of polypeptides, oligonucleotides or lipids based upon the use of phosphoryl, phosphoroamino or phosphorothioate specific hetero-bifunctional linkers combined with fluorescence polarization or fluorogenic detection. Homogeneous assay applications possible using these novel chemistries and methods include in vitro fluorescence polarization assays for the detection of general and isoform specific polypeptide, oligonucleotide and lipid kinase and phosphatase enzymes. Heterogeneous assay applications possible using the same novel chemistries and assay methods enable development of in vitro and in vivo fluorogenic assays of general or specific kinase or phosphatase enzyme activities in cell lysates or serum. In all cases, these new chemistries, methods and assays enable simultaneous multi-kinase detection and profiling.



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# INTERNATIONAL SEARCH REPORT

International application No.

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## A. CLASSIFICATION OF SUBJECT MATTER

IPC(7) : C07K 1/00

US CL : 530/345; 514/2

According to International Patent Classification (IPC) or to both national classification and IPC

## B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)

U.S. : 530/345; 514/2

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practicable, search terms used)  
STN Structure search and East

## C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category *	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	EP 0202758A1 (Imperial Chemical Industries) 14 April 1985 (14.04.85), see example formulas on pages 17-22	1-21
X	US 6,130,323 (Su et al) 10 October, 2000 (10.10.2000), see LC-Biotin-NHSu in Scheme 4 in Figure 5.	22-114

☐ Further documents are listed in the continuation of Box C.

☐ See patent family annex.

\* Special categories of cited documents:

"A" document defining the general state of the art which is not considered to be of particular relevance	"T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention
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"O" document referring to an oral disclosure, use, exhibition or other means	"&" document member of the same patent family
"P" document published prior to the international filing date but later than the priority date claimed	

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